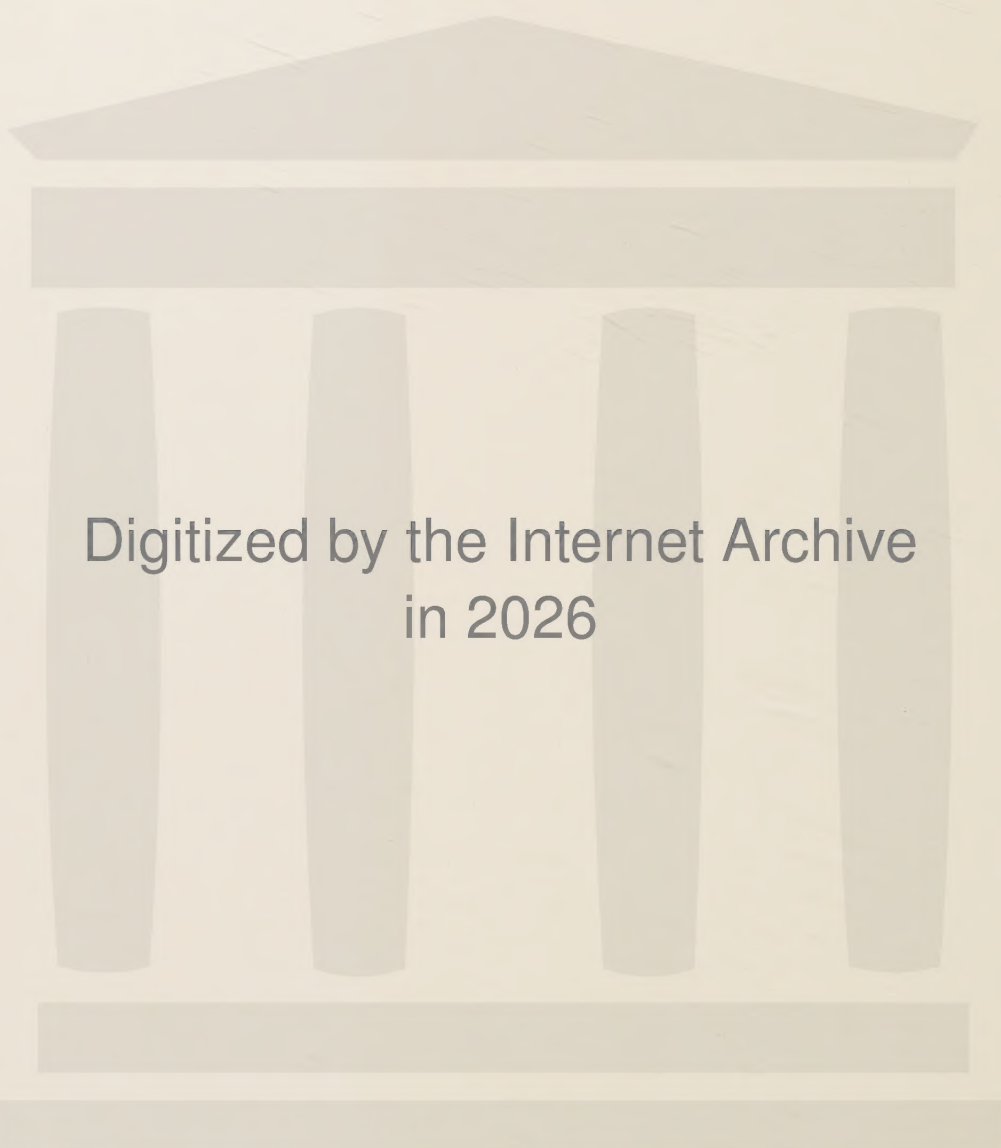


CONTRIBUTION TO THE MASS SPECTROMETRIC STUDY
OF DEUTERIUM-LABELLED FATTY ACID DERIVATIVES

Florence Dinger

Göteborg 1982



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AKADEMISK AVHANDLING

som, för avläggande av filosofie doktorsexamen
vid Göteborgs Universitet,
kommer att offentligen försvaras i
Föreläsningssalen,
Institutionen för Medicinsk och Fysiologisk Kemi,
kl 9.15 lördagen den 8 maj 1982

av

Florence Dinger

This thesis is based on the following reports :

- 1 Andersson, B.Å., Dinger, F. and Đinh-Nguyên, Ng. Organic Syntheses with Heavy Isotopes. II. Methyl vic-dideuterio-*n*-octadecanoate isomers. Chemica Scripta 1981, 18, 197-201.
- 2 Andersson, B.Å., Dinger, F. and Đinh-Nguyên, Ng. Mass Spectrometric Studies of Stable Isotope-labelled Carboxylic Acid Derivatives. I. Normal chain saturated deuterium and carbon-13 compounds. Chemica Scripta 1975, 8, 200-203.
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- 4 Andersson, B.Å., Dinger, F., Đinh-Nguyên, Ng. and Raal, A. Mass Spectrometric Studies of Stable Isotope-labelled Carboxylic Acid Derivatives. III. Methyl perdeuterio-*n*-alkanpyrrolidamates. Chemica Scripta 1976, 10, 114-116.
- 5 Andersson, B.Å., Dinger, F. and Đinh-Nguyên, Ng. Mass Spectrometric Studies of Stable Isotope-labelled Carboxylic Acid Derivatives. IV. Pyrrolidides of vicinal and geminal dideuteriated *n*-octadecanoic acids and the ω -trideuteriated analogue. Chemica Scripta 1982, 19, 118-121.

Abstract

DINGER, Florence, Contribution to the mass spectrometric study of the deuterium labelled fatty acid derivatives, p. 1-17. Department of Medical Biochemistry, University of Göteborg, P O Box 330 31, S-400 33 Göteborg, Sweden (1982).

Mass spectrometry is a well-known powerful aid in the structural determination of lipids. In the contribution to the development of this analytical method several kinds of stable-isotope labelled fatty-acid derivatives have been investigated, so that the fragmentation pathways and the rearrangement mechanisms occurring during electron bombardment could be elucidated. For this purpose the following 80 compounds have been synthesized: the vicinal dideuteriated *n*-octadecanoic acid methyl esters (a whole series of 16 isomers) and the corresponding pyrrolidides (16 isomers), the geminal dideuteriated *N*-*n*-octadecanoyl-pyrrolidines (16 isomers) and an ω -trideuteriated analogue, the *N*-2- ^{13}C -*n*-octadecanoyl-pyrrolidine, the perdeuteriated analogue, the vicinal dideuteriated *n*-octadecenoic acid methyl esters (3 isomers) and the corresponding pyrrolidides (3 isomers), the dimethyl dideuterio-*n*-dodecendioate and the corresponding pyrrolidamate, and the methyl perdeuterio-*n*-alkan ($\text{C}_4\text{-C}_{24}$) pyrrolidamates (21 homologues).

The mass spectrometric analysis of the series of methyl *vic*-dideuterio-*n*-octadecanoates showed that, due to their properties toward electron impact, these esters produced a multitude of fragments and rearrangement ions for different pathways. Thus, it is not easy to determine all positions of the labelled atoms, particularly those which are situated beyond the methylene (11) of the carbon chain.

Under electron impact a pyrrolidide produces a significant series of pyrrolidide-ions and a set of less important miscellaneous hydrocarbon-ions. It is the pyrrolidide-ions that contribute important information about the structure of the carbon chain of the *N*-substituent. All mass spectra of the deuteriated pyrrolidides exhibit the peaks corresponding to these significant ions. In the cases of vicinal and geminal dideuteriated compounds, the mono-deuteriated and dideuteriated ions are easily identified, and the positions of labelled methylenes in the carbon chain are quickly determined.

Due to the charge stabilising effect of the acyl substituent on the nitrogen atom, the methyl pyrrolidamates undergo a regular fragmentation. It is the directing force from the pyrrolidine ring that facilitates, in the case of deuteriated compounds, the location and calculation of isotopic ratio in the carboxylic-acid chain.

The results of the present study shows that the pyrrolidides are, in many aspects, better than the methyl esters for use in the elucidation of structure and the location of labelled atoms of fatty acids by mass spectrometry.

Key words: Mass spectrometry, deuterium label, methyl esters, pyrrolidides, pyrrolidamates.

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The aim of this work was to find the appropriate derivatives of fatty acids for use in mass spectrometric analysis, and to understand the mechanism of fragmentation and rearrangements occurring during electron impact — in the mass spectrometer — in order to facilitate the interpretation of mass spectra of the analysed compounds. These finding and understanding are useful for further biological studies, in which mass spectrometry is a powerful aid for the determination of structure of the metabolites.

Introduction

Application of mass spectrometry to the structural determination of organic long-chain compounds, especially fatty acids, was initiated by Ryhage and Stenhagen et al. [1-14]. Due to the development of combining a gas chromatograph to the mass spectrometer [15-18], mass spectrometry subsequently became the method of choice for the analysis of miscellaneous synthetic and naturally occurring organic substances [19-26].

Concerning fatty acids, the corresponding methyl esters were widely used for mass spectrometric analysis because of their volatility, applicability to gas-liquid chromatography and simplicity of preparation.

Methyl esters of saturated and unsaturated fatty acids yield mass spectra in which the peaks corresponding to the positive ions resulting from the fragmentation and rearrangement of different pathways are exhibited. Mass spectra of the ethylenic esters are complex and do not give information about the primal position of the carbon-carbon double bonds — except that situated at the α -site of the carboxyl group [27] — since, under

electron impact, the ethylenic bonds have a tendency to migrate along the carbon chain [19, 28]. Mass spectra of the acetylenic esters are also as complex [28-30] as those of the preceding esters. Methyl branched chain fatty esters yield mass spectra which, in some cases, it is difficult to elucidate the correct structure of the studied substances [9, 39].

Many attempts have been made in order to determine the position of carbon-carbon double bond(s) in the insaturated fatty esters by mass spectrometry [31-36]. The general method consisted of addition to each ethylenic bond two appropriate atoms or groups of atoms which could be recorded in spectra and could be easily identified. Deuterium-labelling has been used for that purpose, by Đinh-Nguyễn et al. [4, 8, 12] and by Sonneveld [37]. Despite these divers derivatizations, the location of the primal ethylenic bonds did not always give satisfying results.

In order to investigate the mechanism of fragmentation and rearrangement involved in the formation of spectra of the methyl n-octadecanoate, Đinh-Nguyễn synthesized and analysed (by MS) a complete series of geminal dideuteriated methyl n-octadecanoates, an ω -trideuteriated analogue, some vicinal dideuteriated analogues and some $^{13}\text{C}(1)$, $^{13}\text{C}(2)$ and $^{13}\text{C}(3)$ labelled saturated fatty esters [12]. This author reported that, there were difficulties in perceiving the exact position of the deuterium atoms situated beyond the methylene(11) of the carbon chain, because of the extensive fragmentation and rearrangement due to the properties of the methyl esters under electron impact.

Since a tertiary amide, such as N-acyl-pyrrolidine [38],

yields a mass spectrum in which competing fragmentations and rearrangements are minimised due to the charge stabilising effect of the acyl substituent on the nitrogen atom, this amide becomes the derivative of choice for use in the structural determination of divers carboxylic acids possessing: ethylenic bonds [38, 40, 41], acetylenic bonds [42, 43], methyl branching [44-46], oxo group [47], hydroxyl group [47] and cyclopropane ring [47, 55]. The pyrrolidides are, although more polar than the corresponding methyl esters, sufficiently volatile for gas-liquid chromatographic analysis. Depending on the circumstance, the pyrrolidides can be prepared in good yield either from the esters or from the acyl chlorides.

The present thesis concerns:

- a) A completion to the earlier mass spectrometric study [12] of the vicinal dideuteriated methyl n-octadecanoates — report **1**;
- b) A mass spectrometric investigation on some miscellaneous carbon-13 and deuterium labelled N-acyl-pyrrolidines — reports **2**, **3** and **4**;

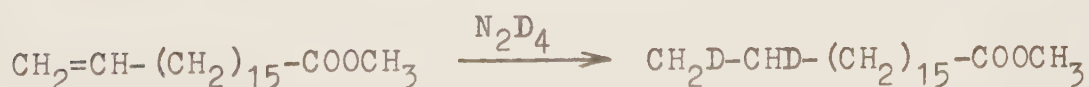
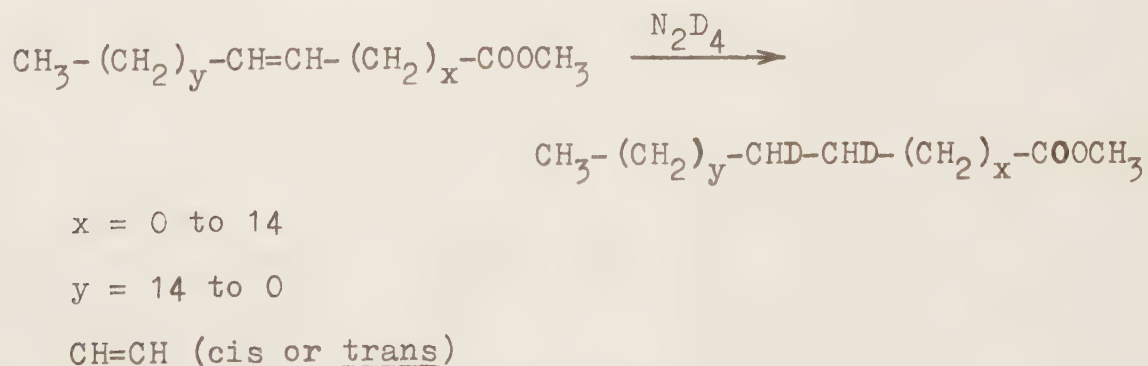
and

- c) A mass spectrometric investigation on two complete series of vicinal and geminal dideuteriated N-n-octadecanoyl-pyrrolidines and an ω -trideuteriated analogue — report **5**.

Synthesis of labelled fatty acids and their derivatives

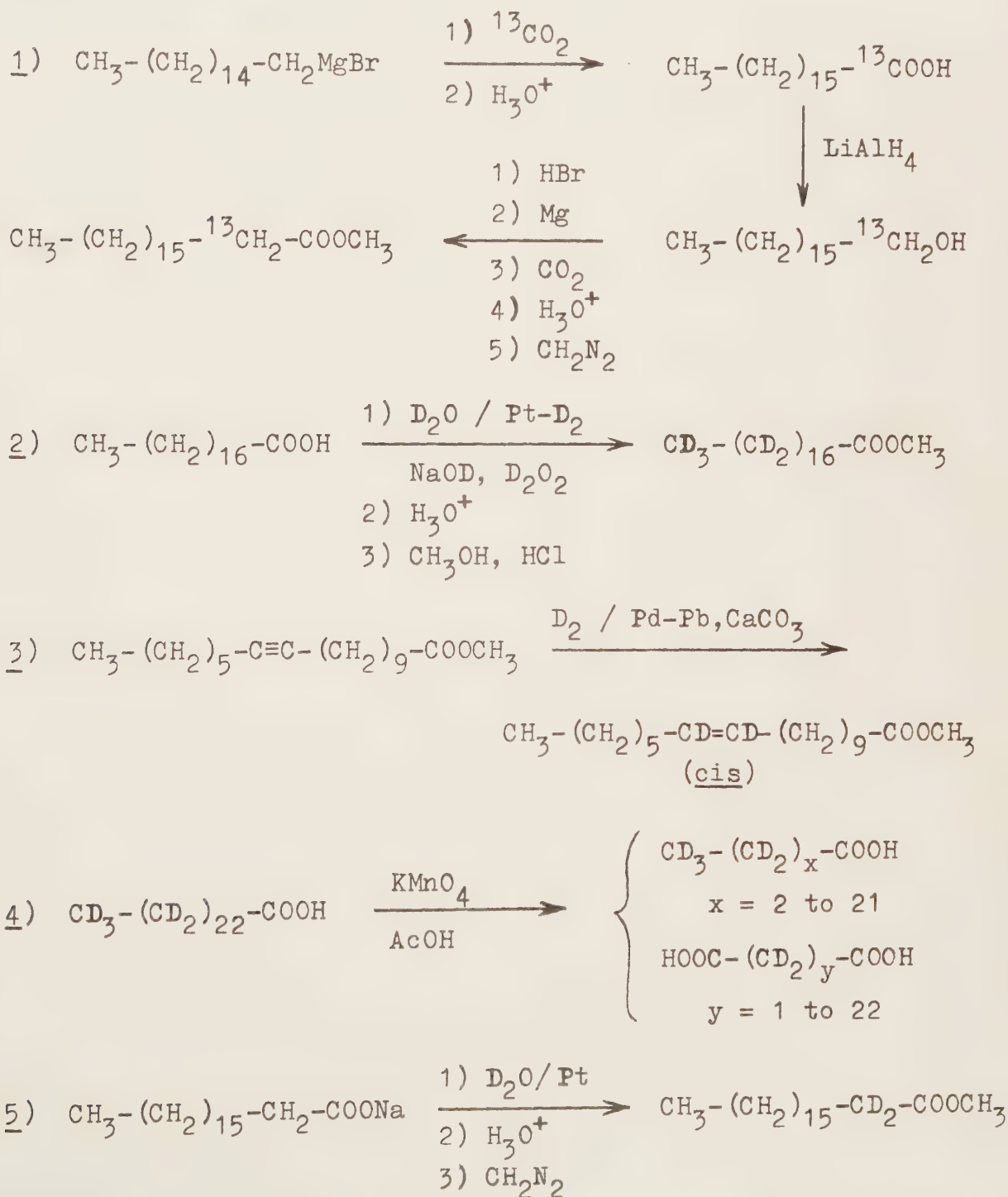
Methods of specifically labelling fatty acids — those subjected in this thesis — with a stable isotope, such as deuterium and carbon-13, have been adjusted and described by Đinh-Nguyễn [12], and a review on deuterium labelling in organic chemistry has been done by Thomas [48].

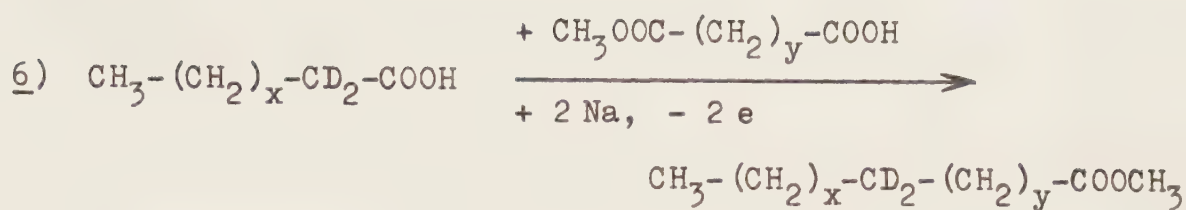
Deuteriation of ethylenic esters using deuterium gas in the presence of a metal catalyst, such as Adams platinum, has been shown to be unspecific because it is often accompanied by cis-trans isomerisation, carbon-carbon double bond migration and protium-deuterium exchange [6, 7, 49]. Reduction of the ethylenic bond involving specific addition of two deuterium atoms has been performed [4, 8, 12] utilising deuterated deuteriohydrazine (D_2N-ND_2, D_2O). The hydrazinic reduction should be executed in anhydrous dioxan in the presence of autoxidised dioxan or oxidised diethyl-ether and air. By using this reduction mode sixteen methyl n-octadecenoate isomers were converted into a complete series of vicinal dideuteriated methyl n-octadecanoates (report 1), as schematised as follows:



Other labelled materials utilised for the preparation of divers pyrrolidides presented in the reports **2-5** — such as

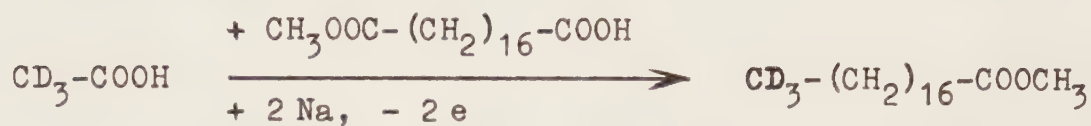
methyl 2- ^{13}C -n-octadecanoate, methyl perdeuterio-n-octadecanoate, methyl 11,12-dideuterio-11c-octadecenoate, dimethyl perdeuterio-n-decandioate, methyl gem-dideuterio-n-octadecanoates (16 isomers) and methyl ω -trideuterio-n-octadecanoate — were synthesized by Đinh-Nguyễn et al. [12, 49-54] and are summarised as follows.





$$x = 14 \text{ to } 0$$

$$y = 1 \text{ to } 15$$



The preparation of pyrrolidides has been performed either from acyl chlorides or from methyl esters, by interaction with pyrrolidine. The aminolysis of the esters should be carried out in the presence of methanol or acetic acid.

Results of the mass spectrometric analysis of the labelled
carboxylic-acid derivatives

Investigation of the labelled carboxylic-acid derivatives by mass spectrometry gave results which are summarised as follows.

In report 1: Sixteen methyl vic-dideuterio-n-octadecanoate isomers have been studied, in order to complete an earlier work by Đinh-Nguyễn [12].

This author predicted fragmentation and rearrangement, and outlined the problem of identifying the exact position of two deuterium atoms from the methylene(11) to the ω -methyl. We could confirm his hypothesis and characterise the feature of the multitude of fragments and rearrangement ions that are involved in the mass spectrum of the methyl n-octadecanoate. Identifying the deuterium labelling close to the ester group gave no difficulty, due to some well-known rearrangement and fragment-ions and the directing effect of the ester group, but in the case of deuterium labelling from methylene(11) to the end of the carbon chain the spectra of the corresponding isomers appear almost identical. Since N-acyl-pyrrolidines have been shown to yield mass spectra which are more simple to interpret, all these esters have been converted into pyrrolidides for further study (see report 5).

In report 2: Several deuterium and carbon-13 labelled N-n-octadecanoyl-pyrrolidines have been investigated.

This shows that the simple fragmentation pattern of the pyrrolidides makes them superior to methyl esters regarding the location of the labelled atoms. It also proves that the reduction by specific addition of deuterium to the ethylenic fatty

acid pyrrolidides seems to be an improved method to locate the carbon-carbon double bonds by mass spectrometry.

In report 3: Three methyl dideuterio-n-octadecanoates, a dimethyl dideuterio-n-dodecendioate, three N-dideuterio-n-octadecenoyl-pyrrolidines and a methyl dideuterio-n-dodecenpyrrolidamate have been investigated.

Identification of the dideuteriated ethylenic bonds is easily performed. This is due to the regular fragmentation which is in accordance with the rule formulated — concerning the non-labelled ethylenic bonds — by Andersson and Holman [40]: If an interval of 12 a.m.u., instead of the regular 14 a.m.u., is observed between the most intense peaks of clusters of fragments containing n and $n-1$ carbon atoms in the acid moiety a double bond occurs between n and $n+1$ carbon atoms in the molecule. All spectra have a distinct molecular ion (M^+), and like earlier described pyrrolidides one dominating fragmentation where the positive charge prefers the pyrrolidide group.

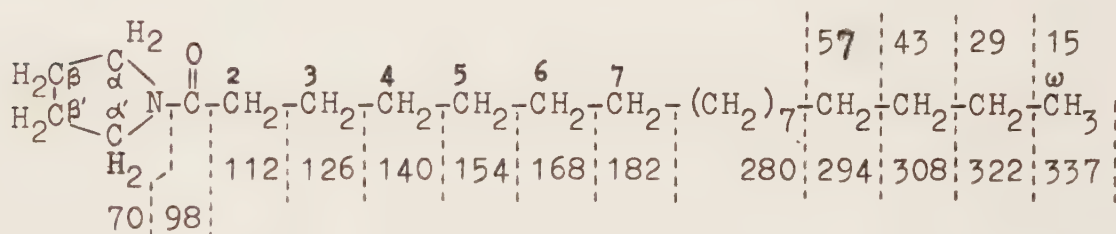
In report 4: Twenty-one methyl perdeuterio-n-alkan(C_4-C_{24})pyrrolidamates and some non-deuteriated analogues have been investigated.

Molecular ion (M^+) of these pyrrolidamates is of much higher intensity than the analogous ion of the corresponding dimethyl esters. The McLafferty rearrangement ion [56], $CH_3OC(\overset{+}{O}D)CD_2$, in the mono and dimethyl esters [54] are usually shown at m/z 77. In the case of pyrrolidides the analogous ion, $C_4H_8NC(\overset{+}{O}D)CD_2$, appears at m/z 116. Due to the charge stabilising effect of the acyl substituent on the nitrogen, the methyl pyrrolidamates undergo a regular fragmentation. It is the direc-

ting force from the pyrrolidine ring that facilitates the location and ratio calculation of isotope in the carboxylic-acid chain.

In report **5**: Sixteen N-vic-dideuterio-n-octadecanoyl-pyrrolidines, sixteen N-gem-dideuterio-n-octadecanoyl-pyrrolidines and an ω -trideuteriated analogue have been investigated.

Under electron bombardment the N-n-octadecanoyl-pyrrolidine produces a significant series of pyrrolidide-ions and a set of less important miscellaneous hydrocarbon-ions. It is the pyrrolidide-ions that contribute important information on the structure of the carbon chain of the N-substituent.



The most intense ion of m/z 113 corresponds to the base peak (100 % relative intensity). This rearrangement ion — presumed as the radical-ion $\text{C}_4\text{H}_8\text{N}^+\text{C}(\text{OH})\text{CH}_2^\bullet$ — is formed probably via a process similar to that postulated by McLafferty [56] for methyl esters. The rearrangement involves the requirement of a hydrogen atom from the methylene(4). This is proved by the fact that the N-4,4-dideuterio-n-octadecanoyl-pyrrolidine yields the analogous ion $(\text{CH}_2)_4\text{N}^+\text{C}(\text{OD})\text{CH}_2^\bullet$ at m/z 114.

The relative abundant ion m/z 126 $\text{C}_4\text{H}_8\text{N}^+\text{C}(\text{OH})\text{CH}=\text{CH}_2$ is accompanied by a less abundant $\text{C}_4\text{H}_8\text{N}^+\text{C}(\text{OH})\text{CH}_2\text{CH}_2^\bullet$ at m/z 127. The mechanism of

formation of these rearrangement ions is comprehended, due to the contribution of the vicinal and geminal dideuteriated pyrrolidides — especially those labelled at the methylenes(2-7). For example, in the case of the N-2,3-dideuterio-n-octadecanoyl-pyrrolidine the fragmentation leading to the rearrangement and for-

mation of $\text{C}_5\text{H}_7\text{N}^+\text{C}(=\text{O})\text{CH}=\text{CHD}$ at m/z 127 and $\text{C}_5\text{H}_7\text{N}^+\text{C}(=\text{O})\text{CH}_2\text{CHD}^+$ at m/z 128 is performed in the first occurrence by successive stereospecific elimination of the deuterium of CHD(2) and capture of a free protium (hydrogen) atom which is lost partly from CH_2 (5), CH_2 (6) or CH_2 (7), and in second instance by capturing another protium coming from any part of the pyrrolidide molecule.

Intensity of the pyrrolidide-ions of type $\left[\text{C}_5\text{H}_7\text{N}^+\text{C}(=\text{O})(\text{CH}_2)_y \right]^+$ (where $y = 3$ to 16) decreases regularly from low to high mass. These ions seem to be formed via the progressive carbon-carbon bond cleavages along the carbon chain. However, the ions of m/z 294 and m/z 308 are formed, partially, by a rearrangement involving consecutive expulsions of a three-carbon fragment and a two-carbon fragment from the carbon chain. This fact is proved by the spectra of 2,3- d_2 , 3,4- d_2 , 2,2- d_2 , 3,3- d_2 and 4,4- d_2 analogues.

The ion corresponding to $M^+ - 1$ is formed by a process entailing abstraction of a protium atom from the molecular ion. This loss may arise from a $\text{CH}_2(\alpha)$ or a $\text{CH}_2(\beta)$ of the pyrrolidine ring.

All spectra of the deuteriated pyrrolidides exhibit the significant ions discussed above. The monodeuteriated and dideuteriated ions are easily identified, and the positions of the labelled methylenes in the carbon chain are quickly determined.

References

1. Ryhage, R., Stenhagen, E. and von Sydow, E., Acta Chem. Scand. 1956, 10, 158; 1957, 11, 180.
2. Asselineau, J., Ryhage, R. and Stenhagen, E., Acta Chem. Scand. 1957, 11, 196.
3. Hallgren, B., Ryhage, R. and Stenhagen, E., Acta Chem. Scand. 1957, 11, 1064.
4. Đinh-Nguyễn, Ng., Acta Chem.Scand. 1958, 12, 1350.
5. Ryhage, R. and Stenhagen, E., Arkiv Kemi 1959, 13, 523; 1959, 14, 483; 1959, 14, 497.
6. Đinh-Nguyễn, Ng. and Ryhage, R., Acta Chem.Scand. 1959, 13, 1032.
7. Đinh-Nguyễn, Ng. and Ryhage, R., J.Res.Inst.Catalysis, Hokkaido Univ. 1960, 8, 73.
8. Đinh-Nguyễn, Ng., Ryhage, R. and Ställberg-Stenhagen, S., Arkiv Kemi 1960, 15, 455.
9. Ryhage, R. and Stenhagen, E., Arkiv Kemi 1960, 15, 291; 1960, 15, 333; 1960, 15, 545.
10. Đinh-Nguyễn, Ng., Ryhage, R., Ställberg-Stenhagen, S. and Stenhagen, E., Arkiv Kemi 1961, 18, 393.
11. Ryhage, R. and Stenhagen, E., in Mass Spectrometry of Organic Ions, ed. F.W.McLafferty, Academic Press, New York, 1963, chap. 9.
12. Đinh-Nguyễn, Ng., Th.Doct., Univ.Paris-Orsay, Ser. A, No. 43, 1967; Arkiv Kemi 1968, 28, 289; Arch.Mass Spectral Data 1971, 2, 100.
13. Stenhagen, E., in Analysis and Characterisation of Oils, Fats and Fat Products, ed. H.A.Boekennoogen, Interscience,

- New York, 1968, vol. 2, chap. 1.
14. Odham, G. and Stenhagen, E., in Biochemical Applications of Mass Spectrometry, ed. G.R.Waller, Wiley-Interscience, New York · London · Sydney · Toronto, 1972, chap. 8.
 15. Ryhage, R., Arkiv Kemi 1962, 20, 185.
 16. Ryhage, R., Anal.Chem. 1964, 36, 759.
 17. Ställberg-Stenhagen, S. and Stenhagen, E., in Topics in Organic Mass Spectrometry, ed. A.L.Burlingame, John Wiley & Sons, 1970, vol. 8, p. 167.
 18. Ryhage, R., Quart.Rev.Biophys. 1973, 6, 311.
 19. Cf. Bieman, K., Mass Spectrometry, McGraw-Hill, New York, 1962.
 20. Cf. Mass Spectrometry of Organic Ions, ed. F.W.McLafferty, Academic Press, New York, 1963.
 21. Cf. Budzikiewicz, H., Djerassi, C. and Williams, D.H., Interpretation of Mass Spectra of Organic Compounds, Holden-Day, San Francisco, 1965.
 22. Cf. Budzikiewicz, H., Djerassi, C. and Williams, D.H., Mass Spectrometry of Organic Compounds, Holden-Day, San Francisco · Cambridge · London · Amsterdam, 1967.
 23. Cf. McCloskey, J.A., in Topics in Lipid Chemistry, ed. F.D.Gunstone, Logos Press Ltd., London, 1970, vol. 1, chap. 6.
 24. Cf. Biochemical Applications of Mass Spectrometry, ed. G.R.Waller, Wiley-Interscience, New York · London · Sydney · Toronto, 1972.
 25. Cf. Spiteller, M. and Spiteller, G., Massenspektrensammlung von Lösungsmitteln, Verunreinigungen, Säulenbelegmaterialien und einfachen aliphatischen Verbindungen,

- Springer-Verlag, Wien and New York, 1973.
26. Cf. Biochemical Applications of Mass Spectrometry, 1st suppl.vol., ed. G.R.Waller and O.C.Dermer, John Wiley & Sons, New York • Chichester • Brisbane • Toronto, 1980.
 27. Ryhage, R., Ställberg-Stenhagen, S. and Srenhagen, E., Arkiv Kemi 1961, 18, 179.
 28. Groff, T.M., Rakoff, H. and Holman, R.T., Arkiv Kemi 1968, 29, 179.
 29. Bohlman, F., Schulman. D., Bethke, H. and Zdero, C., Chem. Ber. 1967, 100, 3706.
 30. Sun, K.K. and Holman, R.T., J.Amer.Oil Chemist's Soc. 1968, 45, 810.
 31. Kenner, G.W. and Stenhagen E., Acta Chem.Scand. 1964, 18, 1551.
 32. Audier, H., Bory, S., Fetizon, M., Longevialle, P. and Toubiana, R., Bull.Soc.chim.France 1964, 3034.
 33. McCloskey, J.A. and McClelland, M.J., J.Amer.Chem.Soc. 1965, 87, 5090.
 34. Niehaus, W.J. and Ryhage, R., Tetrahedron Letters 1967, 5021.
 35. Argoudelis, C.J. and Perkins, E.G., Lipids 1968, 3, 379.
 36. Gunstone, F.D. and Inglis, R.P., Chem.Phys.Lipids 1973, 10, 73.
 37. Sonneveld, W., Ph.D.Thesis, Univ.of Utrecht, 1967.
 38. Vetter, W., Walther, W. and Vecchi, M., Helv.Chim.Acta 1971, 54, 1599.
 39. Jaureguiberry, G., Law, J.H., McCloskey, J.A. and Lederer, E., Biochemistry 1965, 4, 347.
 40. Andersson, B.Å. and Holman, R.T., Lipids 1974, 9, 185.

41. Andersson, B.Å., Christie, W.W. and Holman, R.T., Lipids 1975, 10, 215.
42. Valicenti, A.J., Heimermann, W.H. and Holman, R.T., J.Amer. Oil Chemist's Soc. 1977, 54, 147 A.
43. Valicenti, A.J., Heimermann, W.H. and Holman, R.T., J.Org. Chem. 1979, 44, 1068.
44. Andersson, B.Å. and Holman, R.T., Lipids 1975, 10, 716.
45. Andersson, B.Å. and Bertelsen, O., Chemica Scripta 1975, 8, 91.
46. Bertelsen, O. and Đinh-Nguyễn, Ng., Chemica Scripta (in press).
47. Andersson, B.Å., Progr.Chem.Fats and Lipids 1978, 16, 279.
48. Thomas, A.F., Deuterium Labeling in Organic Chemistry, Appleton-Century-Crofts, New York, 1971.
49. Đinh-Nguyễn, Ng. and Raal, A., J.Res.Inst.Catalysis, Hokkaido Univ. 1969, 17, 171.
50. Đinh-Nguyễn, Ng., Arkiv Kemi 1964, 22, 151.
51. Đinh-Nguyễn, Ng. and Stenhagen, E., Acta Chemica Scand. 1966, 20, 1423.
52. Đinh-Nguyễn, Ng., Raal, A. and Stenhagen, E., Chemica Scripta 1972, 2, 171.
53. Đinh-Nguyễn, Ng. and Vincent, J., Mycopathologia 1976, 58, 137.
54. Đinh-Nguyễn, Ng. and Raal, A., Progr.Chem.Fats and Lipids 1978, 16, 195.
55. Gensler, W.J. and Marshall, J.P., Chem.Phys.Lipids 1977, 19, 128.
56. McLafferty, F.W., Anal.Chem. 1957, 29, 1782.

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